and 50 mM Tris*HCl (pH 7.5) at 37 C. Reaction times varied from 60 min in early rounds to 1 min in later rounds. Each starting pool of templates was based on a sequence complementary to the prototype, with fixed binding arms of seven nucleotides each and a catalytic core randomized to 25% degeneracy at each nucleotide position. For the 8-17 and 10-23 motifs, the templates had the sequence
5'-gtgccaagcttaccgagtaactTCG-TCCGGCTCGGRagatgggtcgtctgtccttccATCT CTAGTTACTTTTTC- 3' and
5'-gttgccaagcttaccg-ggaaaaaTCGTTGTAGCTAGCCtaactaggtcgtctgtccttccA TCTCTAGT TACTTTTTC-3', respectively (PCR primer sites in lower case; substrate-binding arms underlined; randomized positions italicized). The primer used in the template-directed extensions had the sequence
5'-biotin-r(GGAAAAA-GUAACUAGAGAUGG)d(AAGAGATGGCGAC)-3' (SEQ ID NO:132). The PCR primers for the 8-17-based selections were 5'-GTGCCAAGCTTACCGAGTAACT-3' and
5'-d(GGAAGGACAGACGACC-CATC)rU and for the 10-23-based selections were 5'-GTGCCAAGCTTACCGGGAAAAA-3' and
5'-d(GGAAGGACAGACGACCTAGTT)rA. The PCR primers encompassed the binding arms, thus fixing these sequences. One of the PCR primers in each set contained a 3'-terminal ribonucleotide, allowing isolation of the template strand from the double-stranded PCR products by alkaline hydrolysis of the non-template strand and subsequent purification by polyacrylamide gel electrophoresis. A gel-based selection scheme was employed in some of the lineages. In those cases, the PCR primers were
5'-biotin-GTGCCAAGCTTACCG-3' and
5'-GAAAAAGTAACTAG-AGATGGAAGGACAGACGACC-3' and the extension reactions were carried out on the solid support using the primer

5'-r(GGAAAAAGUAACUAGAGAUGGAAG)-3'. A trace amount of [a-32P]-dATP was included in the mixture to label the extension products, which were eluted with alkali, purified by denaturing polyacrylamide gel purification, and recovered by electroelution. The molecules then were reacted and those that underwent cleavage were isolated by gel electrophoresis.

At Page 59, line 26, please substitute the following paragraph for the previous version:

Figure 3 illustrates the sequence alignment of individual variants isolated from the population after five rounds of selection. The fixed substrate domain (5'-GGGACGAATTCTAATACGACTCACTATrAGGAAGAGATGGCGAC-3' (SEQ ID NO:13), or 5'-GGGACGAATTCTAATACGACTCACTATNGGAAGAGATGGCGAC-3', where N represents adenosine ribonucleotide) (SEQ ID NO 13) is shown at the top, with the target riboadenylate identified with an inverted triangle. Substrate nucleotides that are commonly involved in presumed base-pairing interactions are indicated by a vertical bar. Sequences corresponding to the 50 initially-randomized nucleotides are aligned antiparallel to the substrate domain. All of the variants are 3'-terminated by the fixed sequence 5'-CGGTAAGCTTGGCAC-3' (SEQ ID NO 1) ("primer site"; not shown). Nucleotides within the initially-randomized region that are presumed to form base pairs with the substrate domain are indicated on the right and left sides of the Figure; the putative base-pair-forming (or substrate binding) regions of the enzymatic DNA molecules are individually boxed in each sequence shown. The highly-conserved nucleotides within the putative catalytic domain are illustrated in the two boxed

columns.

At Page 61, line 28, please substitute the following paragraph for the previous version:

Synthetic DNAs and DNA analogs were purchased from Operon Technologies. The 19-nucleotide substrate, 5'-pTCACTATrAGGAAGAGATGG-3' (SEQ ID NO:7) (or 5'-pTCACTATNGGAAGAGATGG-3', wherein "N" represents adenosine ribonucleotide) (SEQ ID NO 7), was prepared by reverse-transcriptase catalyzed extension of 5'-pTCACTATrA-3' (SEQ ID NO:8) (or 5'-pTCACTATN-3', wherein "N" represents adenosine ribonucleotide) (SEQ ID NO 8), as previously described (Breaker et al, Biochemistry, 33:11980-11986, 1994), using the template 5'-CCATCTCTTCCTATAGTGAGTCCGGCTGCA-3' (SEQ ID NO 9). Primer 3, 5'-GGGACGAATTCTAATACGACTCACTATrA-3' (SEQ ID NO:6) (or 5'-GGGACGAATTCTAATACGACTCACTATN-3', wherein "N" represents adenosine ribonucleotide) (SEQ ID NO 6), was either 5'-labeled with [γ-^{32}P]ATP and T4 polynucleotide kinase (primer 3a) or 5'-thiophosphorylated with [γ-S]ATP and T4 polynucleotide kinase and subsequently biotinylated with *N*-iodoacetyl-*N'*-biotinylhexylenediamine (primer 3b).

At Page 66, line 2, please substitute the following paragraph for the previous version:

In designing the catalytic domain, we relied heavily on the composition of the most reactive variant, truncating by two nucleotides at the 5' end and 11 nucleotides at the 3' end. The 15 nucleotides that lay between the two template regions were

left unchanged and a single nucleotide was inserted into the 3' template region to form a continuous stretch of nucleotides capable of forming base pairs with the substrate. The substrate was simplified to the sequence 5'-TCACTATrA • GGAAGAGATGG-3' (SEQ ID NO:12) (or 5'-TCACTATN • GGAAGAGATGG-3', wherein "N" represents adenosine ribonucleotide) (SEQ ID NO 12), where the underlined nucleotides correspond to the two regions involved in base pairing with the catalytic DNA molecule.

At Page 17, line 4, please substitute the following paragraph for the previous version:

Figures 4A and 4B illustrate DNA-catalyzed cleavage of an RNA phosphoester in an intermolecular reaction that proceeds with catalytic turnover. Figure 4A is a diagrammatic representation of the complex formed between the 19mer substrate (5'-TCACTATrAGGAAGAGATGG-3', SEQ ID NO 2) and 38mer DNA enzyme (5'-ACACATCTCTGAAGTAGCGCCGCCGTATAGTGACGCTA-3', SEQ ID NO 3). The substrate contains a single adenosine ribonucleotide ("rA", adjacent to the arrow), flanked by deoxyribonucleotides. The synthetic DNA enzyme is a 38-nucleotide portion of the most frequently occurring variant shown in Figure 3. Highly-conserved nucleotides located within the putative catalytic domain are "boxed". As illustrated, one conserved sequence is "AGCG", while another is "CG" (reading in the 5'-3' direction).

In The Claims:

At page 141, line 2, please substitute the following claim 1 for the previously submitted claim 1: